



## Review

# The Antioxidant and Antibacterial Potential of Thyme and Clove Essential Oils for Meat Preservation—An Overview

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**Abstract:** Consumers are looking for safer and more natural food options that are produced through natural methods without using synthetic preservatives. They also desire extended shelf life for their food products. Several medicinal and aromatic plants species combine food, spice, aromatic, and medicinal recognized attributes. The essential oils from these plants contain a unique mixture of compounds specific to each plant, showing notable antioxidant and antimicrobial properties. Essential oils are used widely as they are environmentally friendly, non-toxic, and biodegradable substitutes for harsh chemical preservatives. Thyme and clove are aromatic plants commonly used in traditional gastronomy, particularly in meat-based recipes. The preservation effects of these essential oils on fresh meat have not been widely studied. Therefore, the aim of this study is to review the use of thyme and clove essential oils in meat preservation, with particular emphasis on their antioxidant properties to mitigate lipid and protein oxidation. Different strategies have been used to boost the effects of essential oils in foods, which include mixtures of essential oils, encapsulation and nanoemulsification techniques, with or without edible coatings. The final objective is to promote the wide use of essential oils for meat preservation, eventually in combination with other innovative approaches.

**Keywords:** essential oils; antioxidant activity; antimicrobial activity; meat; thyme; clove



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## 1. Introduction

The rich natural flora of the Mediterranean region has traditionally been inexhaustible used by local populations for centuries. Human and economic development has been dependent on those natural resources. The Mediterranean region is one of the main centers of plant diversity, where around 25,000 species occur, about half of which are endemic to the zone [1]. Moreover, according to Barata et al. [2], the Mediterranean flora is particularly rich in Aromatic and Medicinal Plants (AMP), mainly of the *Labiatae*, *Umbelliferae*, and *Compositae* families, and the region is also considered to be one of the main centers of diversity for AMP.

Aromatic and Medicinal Plants are mostly used for their essential oils (EOs), which are synthesized and stored in special secretory structures located in leaves, flowers, fruits, seeds, barks, and roots. These EOs are volatile at room temperature and may influence both aroma and flavor [3]. These plants have different utilizations: the major use of AMP in the world is as raw materials for essential oil extraction; their non-leafy parts are used as spices for flavoring or seasoning; their leafy or soft flowering parts, herbs, are also used as a flavoring or seasoning; and finally, but not less important, these plants are used,

for example, as medicines, cosmetics, botanical pesticides, herbal drinks/teas, and many other options.

Essential oils are obtained from various AMP typically found in regions with temperate to warm climates, such as the Mediterranean region and tropical areas. Their EOs hold significant value, mainly in traditional medicine, healing practices, and gastronomy. The gastronomy of the Mediterranean region makes use of spontaneous aromatic plants and their essential oils, which have been part of the cultural heritage for centuries.

This review aims to explore the potential for using essential oils as a natural alternative to meat preservation, with a focus on their ability to control lipid and protein oxidation due to antioxidant compounds.

An extensive analysis of the Scopus database was carried out spanning the years 2005 to 2023. The search criteria included articles published in the English language, specifically focusing on research papers that incorporated rigorous experimental designs and appropriate data analysis methods. The following search terms were used: “essential oil”, “natural antioxidant”, “antioxidant properties”, “antioxidant pathway”, “meat”, “preservation”, “thyme”, and “clove”.

In summary, this literature review explores the use of essential oils to preserve meat. The goal is to compile data concerning the application of essential oils in the preservation of meat, with a specific focus on their antioxidant properties in reducing lipid and protein oxidation and antimicrobial properties. The ultimate goal is to encourage the wider utilization of these essential oils for the preservation of meat. This knowledge serves as a valuable resource for informed decision making for the preservation of meats, ultimately reducing food waste without compromising quality and using natural and safe products.

## 2. Essential Oils

According to the European Pharmacopoeia (Ph. Eur.), an essential oil is a fragrant substance typically characterized by a complex composition of secondary metabolites derived from aromatic plants. These oils are obtained through hydrodistillation, steam distillation, or an appropriate mechanical process from a botanically defined raw vegetable material. Primary sources of essential oils include oregano, rosemary, sage, mint, ginger, thyme, and clove, among others [4].

EOs are eco-friendly, biodegradable non-toxic products with potent antimicrobial and antioxidant activities. However, they present limitations when used directly, such as their strong odor and flavor, low stability, low water solubility, high volatility, high degradability, and low bioavailability, which limit their different applications [5].

Nowadays, several active components of EOs are considered as Generally Recognized as Safe (GRAS) by the US Food and Drug Administration (FDA) and are also accepted by the European Commission as flavoring agents in food products [6].

EOs are used in a variety of industrial sectors today, including food processing, pharmaceuticals, perfumes, and cosmetics, and are receiving great interest for their antimicrobial and antioxidant activities [7].

### 2.1. Chemical Composition

Essential oils are typically found concentrated within specific plant cell secretions, primarily situated in various parts of the plant, including secretory trichomes, epidermal cells, internal secretory cells, and secretory pockets. These oils are commonly produced as secondary metabolites and possess antimicrobial and antibiofilm properties, as well as antioxidant activities [8].

These oils consist of a complex mix that can include more than 300 distinct compounds, and they are abundant in substances such as phenylpropanoids and terpenoids [9,10]. The volatile fraction constitutes roughly 90–95% of the entire essential oil, encompassing monoterpenes and sesquiterpene hydrocarbons, along with their oxygenated derivatives. Additionally, it includes aliphatic aldehydes, alcohols, ketones, and esters. In contrast, the nonvolatile residue accounts for approximately 5–10% of the overall oil and is predomi-

nantly composed of hydrocarbons, fatty acids, sterols, carotenoids, waxes, coumarins, and flavonoids [9–13]. The preservative effects of EOs, particularly their antibacterial properties, are predominantly attributed to their phenolic components [14].

The complexity of essential oils comes from their complex chemical composition, with each oil containing approximately 20 to 60 distinct bioactive components. [15]. However, typically, only 2–3 major components are found in relatively high concentrations (ranging from 20% to 70%). Various factors, such as geographic location, environmental conditions, the stage of maturity during harvest, agronomical practices, and the extraction method, can influence the presence and levels of these constituents [15,16]. The choice of the extraction method and the careful optimization of extraction techniques are crucial factors to consider when aiming to produce essential oils with desired characteristics [4].

The antioxidant capacity of essential oils primarily hinges on their chemical compositions. Their effectiveness is closely tied to the existence of phenolic compounds, which possess notable redox properties and play essential roles in counteracting free radicals and breaking down peroxides [15].

The antioxidant effect associated with phenolic compounds stems from their heightened reactivity against peroxy radicals, enabling them to efficiently neutralize these radicals by transferring hydrogen atoms. This observation holds particular significance in the meat industry, as it offers a means of safeguarding meat and meat products from rancidity, ultimately extending their shelf life [4].

Some acknowledged components of essential oils have very pronounced antioxidant and antimicrobial activities and are currently on the market, such as eugenol, thymol, carvacrol, and others [17].

Regarding storage and stabilization of EOs, oxidative and polymerization processes may result in a loss of quality and biological properties, with only a few studies addressing this issue [18]. Nevertheless, this subject has been recently reviewed by Hyldgaard et al. [3] and Naeem et al. [19], and is not the topic of the current work.

## 2.2. Biosynthesis

The biosynthesis of phenolic compounds in plants, starting from aromatic amino acids, begins through the shikimate pathway. The importance of this pathway is underscored by the fact that, under typical growth conditions, approximately 20% of the carbon assimilated by plants follows this route. Within this pathway, aromatic amino acids such as phenylalanine, tyrosine, and tryptophan are synthesized and then either used for protein synthesis or converted through the phenylpropanoid metabolic pathway into secondary metabolites, including phenolic compounds. These phenolic compounds can be categorized into various groups based on the number of carbon atoms comprising their structure and the fundamental phenolic skeleton [14,20].

Essential oil components are chemically categorized based on four criteria: their primary biosynthetic origin; the size or count of carbon atoms; the parent backbone or “skeleton”; and their susceptibility to oxidation by electronegative atoms, such as oxygen, nitrogen, or sulfur, in comparison to carbon atoms. These components can be grouped into three main biosynthetic categories, namely terpenes, phenylpropanoids, or isothiocyanates, originating from four major biosynthetic pathways: the mevalonate and methylerythritol phosphate pathways, the shikimate pathway, and the glucosinolate pathway [21].

Two different biochemical pathways, namely the mevalonic acid pathway, active in all eukaryotes, and the 2C-methyl-D-erythritol-4-phosphate pathway, restricted to higher plants, generate the precursors of isoprene, isopentenyl, and dimethyl-allyl diphosphate [22–24].

The shikimate pathway yields several compounds, including phenols, phloroglucinols, lignin, flavonoids, and amino acids, among others, and is responsible for the synthesis of phenylpropanoids [21,25–27].

Finally, isothiocyanates are synthesized via the glucosinolate pathway [21].

### 2.3. Protective Effect

Essential oils have been widely studied due to both their antimicrobial and antioxidant activities. In the context of human health concerns, the United States Environmental Protection Agency has validated the effectiveness of certain EOs and, consequently, registered citronella, lemon, and eucalyptus oils as ingredients for insect repellents applied to the skin. These natural products are increasingly favored due to their proven low toxicity, comparable efficacy when compared to other EOs and synthetic chemicals, and their popularity among consumers [14].

#### 2.3.1. Antimicrobial Activity

The antimicrobial attributes of EOs are attributed to a variety of compounds, broadly categorized into terpene and phenolic compounds. Essential oils exhibit antimicrobial efficacy against a broad spectrum of microorganisms; however, the precise antimicrobial mechanism remains incompletely understood, and it cannot be attributed to a single mechanism [10,15,28]. The biological effects of EOs are dictated by the composition, concentration, and proportions of their chemical constituents [29]. It appears that, based on the chemical constituents present in essential oils, several mechanisms contribute simultaneously to their antimicrobial properties. In general, the interaction between EOs and bacterial cell membranes can be effective in impeding bacterial growth. Factors such as the hydrophilic or lipophilic properties of EO constituents, the type of microorganism, and the structure of the cell wall influence the antimicrobial activity of EOs. The mechanisms of antimicrobial action of EOs vary based not only on the primary chemical composition, but also on whether the action is directed at Gram-positive or Gram-negative bacteria or fungi commonly found in foods, including meat [16]. Gram-positive bacteria tend to be more susceptible to EOs than Gram-negative ones [8].

Initially, EOs disrupt cellular structures, compromising membrane integrity, increasing permeability, and interfering with cellular activities such as energy production and membrane transport. This membrane disruption leads to alterations in vital processes such as nutrient processing, synthesis of structural macromolecules, and growth regulators. Due to their lipophilic nature, EOs easily penetrate bacterial cell membranes, causing leakage of cellular components and ion loss [16,30]. In bacteria, the disintegration of the plasma membrane results in the loss of RNA, proteins, and ions, disrupting the delicate ionic gradient crucial for cellular survival. Potassium, vital for maintaining osmotic balance, turgor pressure, pH, glucose metabolism, oxygen consumption, and enzyme activation, is depleted through the disruption of cell membranes by EOs, leading to cell death [29]. This susceptibility disparity may stem from the rigid outer membrane rich in lipopolysaccharide present in Gram-negative bacteria, which is hydrophobic and restricts the diffusion of hydrophobic EO compounds. In contrast, the less dense peptidoglycan layer of Gram-positive bacteria allows small antimicrobial molecules, particularly small lipophilic compounds, to seamlessly integrate and traverse the phospholipid bilayer [31]. This integration damages the cell membrane structure, compromising its functionality. Substantial leakage of cell content, ions, and essential molecules ultimately culminates in cell death [28].

Membrane proteins are essential for undisturbed membrane functions, and certain EO compounds have been identified to denature these proteins, thereby impairing membrane functions or influencing the synthesis of the cell wall structure. Some EO compounds have shown the ability to block enzyme functions, hindering metabolic pathways, binding to DNA, or impacting protein synthesis [31]. Additionally, EOs exert antibacterial activity through various means, including altering cell membrane fatty acid profiles, compromising cytoplasmic membrane integrity, inducing cytoplasmic coagulation, depleting proton motive force, influencing adenosine triphosphate (ATP) synthesis and hydrolysis, reducing membrane potential, suppressing quorum sensing, and hindering bacterial biofilm formation and virulence factor expression [30].

The antibacterial effect of essential oils and other plant extracts results from combined mechanisms of action, such as: the antibacterial activity of phenolic compounds; the

changes in the permeability of microbial cells; and the interference with cell membranes, altering their structure and function [28].

The antifungal actions of EOs mirror antibacterial mechanisms through direct contact with fungi. However, EOs also exhibit antifungal activity in the vapor phase, particularly against molds. On contact, EOs penetrate and alter the fungal cell wall and cytoplasmic membranes through a permeabilization process, disintegrating mitochondrial membranes. In the vapor phase, EOs disrupt the life cycle of some molds during germination, affecting hyphal growth and sporulation. Inactivation of conidia by EOs is crucial, considering conidia's stability to heat, light, and chemical compounds, making them challenging to eliminate [16]. EOs with higher concentrations of terpenoids demonstrate enhanced antifungal activity compared to those rich in monoterpenes and sesquiterpenes [29].

### 2.3.2. Antioxidant Activity

Essential oils, as natural antioxidants, employ multiple mechanisms to decelerate oxidation processes. They act by impeding the initiation and progression of chain reactions, scavenging, and terminating free radicals, quenching the formation of singlet oxygen, and binding to transition metal ions that serve as catalysts in oxidation reactions. Phenolic compounds are a dominant presence in many essential oils, comprising up to 85% of the total composition in some cases [14,15,21]. These substances act as key antioxidants, disrupting oxidation chains, and effectively scavenging free radicals. They play a crucial role in preventing lipid oxidation, hindering the initial phases of oxidation reactions, and maintaining the quality of meat and meat products. Moreover, oxidative processes can affect proteins, with lipid-derived reactive oxygen species, such as peroxy radicals, having the capacity to initiate protein carbonylation. Phenolic compounds contribute to inhibiting these oxidation reactions by slowing down lipid oxidation, establishing complexes with proteins, and thereby alleviating oxidative damage [4,32].

The presence of saturated fats in meat makes it prone to oxidative degradation when exposed to air, resulting in the development of off flavors. Oxidative deterioration is exacerbated by prooxidants such as free iron. Phenolic compounds in EOs contribute antioxidant properties, effectively preserving meat against lipid peroxidation. However, some EOs may exhibit prooxidant cytotoxic properties in eukaryotic cells by damaging mitochondrial membranes. EO phenols, can switch from an antioxidant to a prooxidant state at high concentrations, damaging mitochondrial membranes, releasing reactive species, and potentially harming DNA, proteins, and other cellular components [29].

Reducing lipid oxidation poses a significant challenge in meat processing, cooking, and refrigerated storage, as it leads to the generation of harmful compounds that diminish the nutritional quality of meat and its derivatives. This oxidation process is driven by a chain reaction initiated by peroxy radicals ( $\text{ROO}\bullet$ ). Essential oils, which act as natural antioxidants, prevent or retard lipid oxidation in meat and meat products [4]. Nevertheless, owing to the diverse array of active compounds present in EOs, their antioxidant activity cannot be ascribed to a singular mechanism. The plausible mechanisms through which EOs inhibit oxidation reactions encompass averting the initiation of chain reactions, halting the ongoing extraction of hydrogen, scavenging free radicals, hindering the formation of singlet oxygen, and binding to catalysts composed of transition metal ions [30].

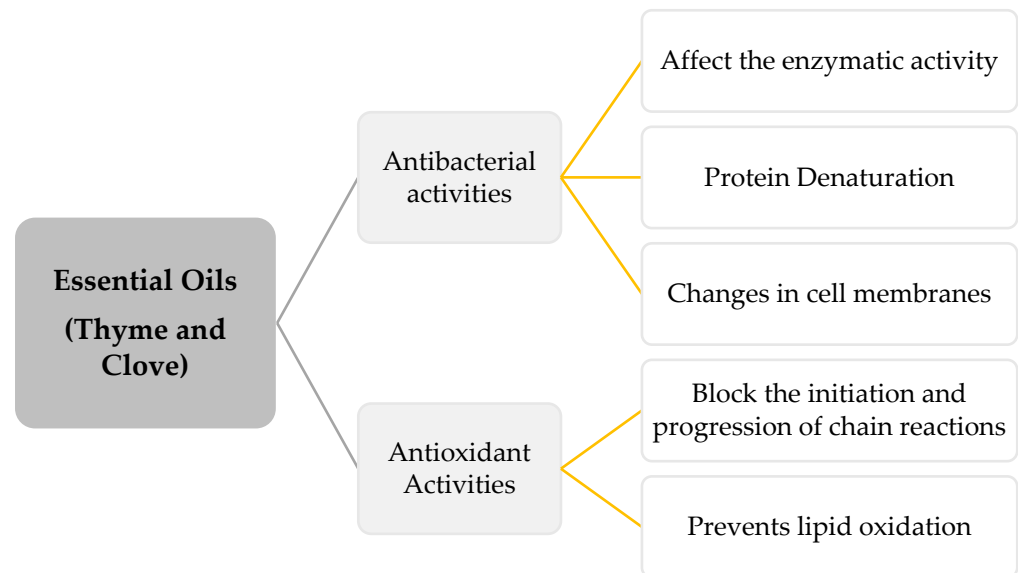
Among these compounds, phenolic substances, such as thymol, stand out as the most well-known natural antioxidants, recognized for their role as classic chain-breaking antioxidants [4]. The hydroxyl group ( $-\text{OH}$ ) present in phenolic compounds acts as a site for hydrogen atom donation, participating in reactions with peroxy radicals generated during the oxidative breakdown of unsaturated fatty acids. This interaction results in phenoxy radicals which are unable to extract hydrogen atoms from unsaturated fatty acids. Phenoxy radicals engage with lipid radicals, leading to the creation of non-radical species, the deactivation of peroxy radicals, and the end of oxidation reactions. Phenolic compounds have the ability to inhibit oxidation reactions by slowing down lipid oxidation, binding to proteins, and forming complexes with them [30].



#### 2.4. Thyme and Clove Essential Oils

Recently, several studies have been conducted to understand the importance of using essential oils in food preservation, mainly meat and meat products, with special emphasis on thyme and clove essential oils [13,33,34].

Thyme and clove essential oils have several mechanisms of antibacterial and antioxidant activities, which are summarized in Figure 1.



**Figure 1.** Mechanisms of antibacterial and antioxidant activities of essential oils.

##### 2.4.1. Thyme (*Thymus vulgaris* L.)

*Thymus vulgaris* L. is a perennial aromatic woody plant belonging to the *Lamiaceae* family and the genus *Thymus* L., that includes around 215 species of subshrubs and herbaceous perennials [35,36]. Other authors pointed out a higher number for Genus *Thymus*, stating that it comprises a total of 928 species [37].

Thyme is a small bush, reaching no more than 40 cm high. Normally, leaves are used for the EO extraction. Flowers are axillary and grouped at the tip of the branches (Figure 2) [38].



**Figure 2.** Thyme plant (*Thymus vulgaris* L.).

This genus is native to Southern Europe, the western Mediterranean Basin, but *T. vulgaris* L. has a worldwide distribution [39]. These plants are well adapted to the arid climate in the Mediterranean Basin, in sunny, warm–temperate climate conditions and are resistant to

frost. A situation of prolonged dry periods is supported by thyme, but, on the other hand, is important that soils are well drained [38].

The thyme EO contains more than 60 compounds, however, considering a practical point of view, the major part and most active compounds are thymol (68.1%) and carvacrol (10%) [40]. Galovićová et al. [41] studied the main components of thyme EO and found that they were thymol (48.1%), *p*-cymene (11.7%), 1,8-cineole (6.7%),  $\gamma$ -terpinene (6.1%), and carvacrol (5.5%). Thymol is well known for its antimicrobial and antiseptic properties, carvacrol contributes to the sharp, spicy fragrance, with strong antimicrobial properties, and *p*-cymene possesses antioxidant properties and is being studied for its potential role in protecting against oxidative stress [20,42,43]. According to Galovićová et al. [41], there are six known chemotypes of *T. vulgaris* essential oils: geraniol, linalool,  $\alpha$ -terpineol, tujanol-4, thymol, and carvacrol.

*T. vulgaris* L. has been used in traditional medicine for many centuries due to the antimicrobial properties as well as powerful antioxidant properties [42,44–46].

The role of *Thymus vulgaris* L. EO as a preservative for food products is explained by: its activity as an antioxidant, its antimicrobiological properties, its ability to control lipid oxidation, and its ability to increase stability during the shelf life.

The essential oils of thyme have proven efficacious against bacteria as well as yeasts, and even against filamentous microscopic fungi [47]. The recognition of the effect of thyme EO on the preservation of food products, due to its antioxidant, antimicrobial, and lipid degradation-lowering properties, led to it being selected as a reference in the studies carried out by Sacchetti et al. [48] with oils from 11 different AMP.

In addition, the wide availability, pleasant taste and smell, and the fact that it is often used in cooking recipes as a flavoring agent in traditional food by many consumers are all positive factors.

#### 2.4.2. Clove (*Syzygium aromaticum* L.)

*Syzygium aromaticum* L. is a native plant from the Moluccas islands in Indonesia, but is widespread and cultivated in tropical and subtropical regions such as Madagascar, Sri Lanka, and China. *Syzygium aromaticum* L. belongs to the *Myrtaceae* family, which has more than 3000 species and up to 150 genera [49].

The commercial parts of the tree are essentially the buds, and production begins four years after plantation. This is an evergreen tree that can reach a height of 12 m, with reddish flowers organized in terminal clusters. The flower buds exhibit a bright red color, and when they reach 1.5 to 2.0 cm are ready for collection. They are composed of a long calyx, four spreading sepals, four unified unopened petals forming a small central sphere (Figure 3) [50,51]. Most oils are extracted from the vegetative parts of the plants, but this is an exception. The aromatic dried flower buds of the tree *Syzygium aromaticum* L. are called cloves and are the raw material for EO extraction. Very seldom are the leaves used.



**Figure 3.** Dried flower buds of the clove tree (*Syzygium aromaticum* L.).

Considering the study conducted by Jirovetz and colleagues [52], cloves comprises a total of 23 identified constituents, among them eugenol (76.8%), followed by  $\beta$ -caryophyllene (17.4%),  $\alpha$ -humulene (2.1%), eugenyl acetate (1.2%), and methyl salicylate. Eugenol is a phenolic compound that contributes to clove oil's sweet and spicy aroma, with antibacterial and analgesic properties, caryophyllene is a sesquiterpene with woody undertones and anti-inflammatory properties, eugenyl acetate contributes to the sweet, fruity notes and offers some antimicrobial properties, and methyl salicylate imparts a mild minty aroma with analgesic and anti-inflammatory properties [53–55]. Clove flower buds contain up to 18% of essential oil [52].

According to Batiha et al. [56], the major constituents of clove EO are carvacrol, eugenol, thymol, and cinnamaldehyde. Eugenol is the clove EO active substance. As for all the other AMP, their composition changes according to several factors. It is well known that the phenological stage influences the yield and quality of clove essential oil and, according to Alfikri et al. [57], the clove flower buds in the flowering stage had the highest yield, eugenol content, and refractive index. They also state that buds of mature trees gave the best quality of clove EO, while the one obtained from young trees had the strongest antioxidant activity.

Clove EO richness in volatile compounds and its antimicrobial and antioxidant activities have led to its widespread use as a flavoring in gastronomy and in food industries, but it also has other uses in health, medical, and cosmetic products. For a long time, various authors such as Chomchalow [58] and Cortez-Rojas et al. [49] have considered it to be very promising for preserving food products, with relevance for meat products, due to their antioxidant and specially antimicrobial properties. In addition to this, the fact that the FDA considers clove to be GRAS should be considered.

Moreira et al. [59] studied the effect of clove EO in vitro and determined the minimal inhibitory concentrations and optimal contact time between pathogen and EO. Later on, Moreira et al. [60] evaluated the effectiveness of clove to control *Escherichia coli* O157:H7 at real food conditions (8–10 °C and 20–22 °C temperatures). They found that oil concentration, food composition, and storage temperature were determinant for the antimicrobial real action of clove EO. Clove EO exhibited a bactericidal and bacteriostatic action on the survival and growth of *E. coli* O157:H7. The relative concentration of the main compounds is decisive for the antioxidant and antimicrobial properties of clove [61,62].

A recent study by Battagin et al. [63] emphasizes the role of EOs as a substitute for traditionally used chemical agents. For citrus fruits marketed in Europe, they could not be sanitized with sodium hypochlorite due to a prohibition from the European Commission. Therefore, in this case study, its substitution by clove EO was evaluated. Tests in vitro and in vivo, and the simulation of the sanitization process in packinghouses, showed that the application of clove EO had the same sanitization efficacy as NaOCl considering the presence of *Xanthomonas citri* subsp. *citri* (*X. citri*), as was originally intended.

### 3. Use of Essential Oils in Meat Preservation

Meat is a fundamental component in diets around the world, extremely important for human nutrition and a vital source of nutrients [64].

However, meat is highly susceptible to biochemical and microbial deterioration, particularly during preservation, due to the presence of saturated and unsaturated lipids, proteins, carbohydrates, vitamins, minerals, and heme pigments [4,64,65]. According to Kumar and colleagues [66], high concentrations of unsaturated fatty acids favors meat oxidation.

Meat is susceptible to spoilage by bacteria (e.g., *Pseudomonas*, *Acinetobacter-Moraxella*, Enterobacteriaceae), molds (such as *Fusarium*, and *Mucor*), and yeasts, namely *Candida* spp.

Fresh meat may undergo oxidative reactions upon exposure to atmospheric oxygen, UV radiation, endogenous enzymes, free radicals, and transition metals. These reactions produce chemical compounds that have cytotoxic, mutagenic, and oxidative effects on human tissues, accelerating aging processes and eventually causing cancer, atherosclerosis, and inflammation [67,68].



Oxidative reactions occur during the conversion of muscle into meat, meat processing, and storage, and constitute one of the main causes associated with the degradation of meat quality [4,69]. The oxidative process induces the degradation of lipid, protein, and color pigment, which leads to losses of nutritional value, texture changes, development of an off flavor and off odor, perceptions of rancidity, discoloration, and the development of toxic compounds, resulting in reduced shelf life [4,69–73].

It should be noted that this type of change occurs more quickly in minced meat than intact meat, because minced meat has a larger surface area which facilitates direct interaction between lipids and air and provides better accessibility for oxidation promoters. These promoters include heme and non-heme iron from meat pigments such as hemoglobin, myoglobin, and phospholipids from disrupted cells [73,74].

The oxidative stability of meat depends on several intrinsic and extrinsic factors, including enzymatic activity, pH, pro-oxidants concentration, temperature, and protein and lipid fraction compositions, which vary according to the animal species [69,75].

Lipids, proteins, and pigment oxidation regularly occur in meat and meat products under conventional storage conditions [4]. According to Zahid and coworkers [73], frozen storage of meat is considered the most effective procedure to prevent oxidation, allowing the preservation of the meat's quality attributes for a longer period.

To increase the shelf life of meat, the industry has been using synthetic preservatives to prevent or slow down oxidative reactions. However, the majority of these compounds are of synthetic origin, with high concentrations being used, and it is recognized that they negatively affect consumer health due to their toxicological and carcinogenic effects and consumers are aware of this [30,69,76]. The most common food antioxidant additives are butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), propyl gallate (PG), and tert-butylhydroquinone (TBHQ) [30,69].

In recent years, consumers have changed their consumption habits, and keep looking for new food options that combine natural and biologically active ingredients that promote health and are free from additives. The demand for more healthy and eco-friendly products is currently one of the biggest challenges for the food industry. The food market is looking for natural antioxidants. These antioxidants can prevent oxidative chain reactions, diminish oxidation processes, and extend the shelf life of high-fat meat [30,66]. Thus, the use of several plant extracts and essential oils from different sources, such as aromatic plants, fruits, leaves, seeds, and spices, has emerged as a viable alternative to preserve meat and reduce or delay spoilage [11,68,77].

The effect of natural antioxidants in meat varies with the composition of the plant extract, their antioxidant activity, application form, food processing, and meat matrix considering the fraction compositions of the lipid and protein [69].

It is also important to note that extracts and essential oils have different biological activities according to the nature of the raw material and the obtention process, so it is extremely important to have a good assessment of the chemical composition due to their complex natural mixtures and often synergistic effects with other components [78].

According to Cunha and coworkers [69], the conventional method of incorporating natural antioxidants into meat (pieces, slices, ground or small pieces) is more effective at preventing lipid and protein oxidation than spraying them onto the surface, because there is more contact between susceptible compounds and antioxidants. However, this application method has been subject of some controversy, considering that compounds based on botanical sources have low water solubility, low chemical stability, and limited biological activity, which could be a problem when acting as antioxidants [79]. In recent years, several studies have been carried out on meat and meat products to evaluate the effectiveness of essential oils as antimicrobials and antioxidants (Table 1).

**Table 1.** Impact of different preservation protocols on the shelf life of different meats and meat products.

| Foods               | Treatments  | Experimental Conditions  | References |
|---------------------|---|--|------------|
| Porcine liver pâté  | Sage and Rosemary EOs and BHT synthetic antioxidant | Refrigerated storage at 4 °C/90 days   | [80]       |
| Cooked ground beef  | Clove extract and BHT synthetic antioxidant         | Cooked ground beef during frozen storage for 6 months at freezing (−20 °C). 0.1% clove extract and 0.02% BHT.  | [73]       |
| Fresh pork meat     | Oregano and thyme EOs                               | Refrigerated storage at 2 °C/6 days. Impregnated cotton gauze with oregano and thyme EOs (diluted 1:10 in water).  | [13]       |
| Chicken breast meat | Thyme and balm EOs                                  | Refrigerated storage at 4 °C/3 weeks. 0.5% thyme and balm Eos.   | [81]       |
| Raw beef meat       | Thyme EO, acetic acid, and combination              | Refrigerated storage at 4 °C/15 days. 2% thyme essential oil, 2% acetic acid, and 2% mixture of thyme EO and acetic acid.  | [82]       |
| Ground beef         | Clove and cinnamon EOs                              | Refrigerated storage at 8 °C/7 days, chilling stored at 0 °C/7 days, and freezing stored at −18 °C/60 days. 5% and 10% of crude and commercial clove EO and 2.5% and 5% of crude and commercial cinnamon EO. | [83]       |
| Minced beef         | Thyme and clove EOs                                 | Refrigerated storage at 4 °C/9 days. 2 and 4 MIC (Minimum inhibitory concentration) for thyme and clove Eos.   | [78]       |

Estévez and colleagues [80] studied the impact of synthetic and natural antioxidants on protein oxidation in porcine liver pâté, finding evidence that the use of sage and rosemary essential oils exhibited similar antioxidant properties to BHT, therefore suggesting that they could be a viable alternative to synthetic antioxidants. More recently, Zahid and colleagues [73], considering the utilization of *Syzygium aromaticum* extract as a natural antioxidant instead of BHT in cooked ground beef during frozen storage, found that lipid oxidation and volatiles formation were minimized.

According to Laranjo and co-workers [13], the application of thyme EO in pork meat showed promising results in reducing the count of microbial groups, particularly for the population of enterobacteria. However, oregano EO revealed no significant reduction in the population of the studied microbial groups.

Fратиanni et al. [81] evaluated the effect of thyme essential oil on the preservation of chicken breast meat during a storage period of 3 weeks at 4 °C. The results showed that the application of 0.5% of thyme essential oil is effective in preserving the meat. Throughout the storage period, radical formation is reduced, lipid peroxidation is decreased, sarcoplasmic proteins are preserved, deterioration reactions are minimized, and the shelf life of chicken breast meat is extended. The thyme EO exhibited a DPPH (2,2-diphenyl-1-picrylhydrazyl) inhibition percentage ranging between 25% and 30% when compared to the control treatment. It is important to state that the higher antioxidant activity of thyme EO is due to its high carvacrol content. Additionally, the application of this oil led to a decrease in the natural microflora present in the meat, and effectively inhibited the growth of *E. coli*.

Asuoty and co-workers [82] obtained similar results in their study by applying thyme essential oil and acetic acid to fresh beef steaks. The results indicate that treating raw beef meat with thyme EO, acetic acid, and their combination has a beneficial effect on reducing meat oxidation compared to the control treatment, allowing the shelf life of this product to be extended.

The study of Khaleque and colleagues [83] showed that 10% clove EO completely inactivated *Listeria monocytogenes* after 3 days at storage temperature, however, cinnamon EO was not effective in inactivating.

The in vitro study carried out by Zengin and Baysal [78] has shown that clove essential oil had greater antioxidant activity than thyme oil. In the same study, the authors evaluated the effect of these two essential oils in controlling the lipid oxidation of minced beef, and observed that the EOs were able to delay lipid oxidation during 9 days of storage at 4 °C. The essential oils of thyme and clove showed strong antimicrobial activity against all the spoilage and pathogenic bacteria tested.

Recent publications have considered the application of plant extracts and essential oils in meats through the use of encapsulation and nanoemulsification techniques [84], as well as their inclusion in the formulation of edible coatings and active packaging [68]. Thus, their dispersibility, chemical stability, and matrix compatibility can be guaranteed [79]. These new methods improve the cost–benefit ratio of adding natural antioxidants to meat by reducing the amount of antioxidant used and allowing these compounds to be more safely and cleanly applied to food [68,84]. Through these technologies the natural antioxidant is applied to the intrinsic component of the packaging and polymer film, and so the antioxidant effect gradually acts on the packaged meat [68,79].

Obtaining natural antioxidants could involve the use of by-products from the food industry, from a circular economy perspective, allowing recovering waste from various industries [79]. Free-form EOs engage with lipids in microbial and mitochondrial membranes, increasing permeability, inducing changes in membrane potential, leading to ion loss, disrupting the proton pump, and disturbing the microbial metabolism, ultimately resulting in lysis and microbial death. In contrast, the precise mode of action of nanoencapsulated EOs is not fully understood, with hypothesized enhancement of activity due to reduced size facilitating more efficient interaction with cell membranes. Lower doses can be employed, and certain carriers possess antimicrobial activity, altering membrane potential, generating reactive oxygen species, and influencing microbial metabolism. The antimicrobial efficacy of EOs is primarily attributed to phenolic components, with a suggested synergistic effect involving minor components such as monoterpene hydrocarbons. Additionally, some propose synergistic actions between nano-EOs and encapsulation carriers, shielding EOs from food matrices and facilitating transfer to specific targeted sites [85]. According to Šojić and colleagues [86], the use of wild thyme by-product extract is useful as a natural antioxidant in ground pork patties, because it inhibited lipid oxidation and reduced protein oxidation during storage (4 °C for 3 days).

Additionally, the incorporation of natural antioxidants into animal feed is a good way to guarantee the oxidative stability of meat. Many studies have shown the benefits of adding natural antioxidants (e.g., thyme, clove, oregano, and rosemary) to animal feed, with effects on the animal's health and performance, as well as on the quality of the meat [87–90].

The incorporation of essential oils into food formulations can have an impact on sensory quality and consumer acceptance [84]. According to Danilović et al. [91], the addition of essential oils to minced pork, especially in high concentrations, in order to control oxidative stability and microbial growth, can cause changes in the sensory characteristics of the product, particularly due to the intense smell and flavor. It is therefore important to choose minimum concentrations of essential oils, to ensure that the concentration is sufficient for their antioxidant action, antimicrobial activity, and to increase the shelf life, but without sensory alterations that depreciate the final product.

#### 4. Conclusions

This review confirms the importance of using EOs as substitutes for synthetic food preservatives. Consumers are looking for safer, natural foods, free from synthetic preservatives, but they also want a long shelf life for food products.

Meat and meat products are predisposed to oxidation and microbial contamination, causing undesirable changes in sensory and nutritional characteristics, and increasing the

risks regarding food safety. Consequently, the increase in food loss and waste with the consequent economic damages are felt by all stakeholders and the use of EOs is seen as an interesting alternative to chemical preservatives.

EOs are used in the food industry to enhance flavor and for the preservation of foods thanks to the antimicrobial and antioxidant effects reducing the use of synthetic preservatives and increasing the shelf life. Thyme and clove EOs have antimicrobial and antioxidant activities with good results in the control of oxidation reactions, allowing for the preservation of meat. In addition, their flavors are welcomed by consumers who are already used to them.

Innovative approaches are being used, such as combinations of different EOs, use of encapsulation, and nanoemulsification techniques in association with edible coatings, as a strategy to increase the synergistic and additive effects of EOs in foods.

Nevertheless, more research studies are needed to highlight the most adequate strategy to effectively preserve meat products throughout the whole desirable shelf-life period without compromising the product acceptability for consumers.

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